

SOUND PRODUCTION AND RECEPTION IN THE
CARIBFLY, *ANASTREPHA SUSPENS*A
(DIPTERA: TEPHRITIDAE)

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ABSTRACT

Virgin female *Anastrepha suspensa* (Loew) with aristae removed failed to respond to recorded male "calling" song to the same extent as intact females. However, removal of aristae did not change female response to another sound known to modify behavior: the "precopulatory" song sung by mounted males. Thus, aristae are possibly not the sole organs of sound/vibration reception. Virgin females without aristae are attracted to male-produced pheromones and the aristae bear no evidence of chemosensilla. These organs appear to be involved only in mechanoreception. Apparently the male songs of *A. suspensa* are produced solely by wings because their sound pressure levels increase in proportion to the amount of wing surface, and there are no obvious morphological features that might make up a wing-powered stridulatory mechanism. There is a slight sexual dimorphism in wing shape (male wings were more oval). Comparisons among tephritids with various wing shapes and courtship signals suggest that the dimorphism might be due to male wing modification for sound production.

RESUMEN

Hembras vírgenes de *Anastrepha suspensa* (Loew) con las aristas removidas, no respondieron en el mismo grado que las hembras intactas al canto grabado de llamada del macho. Sin embargo, al quitarle las aristas, no cambió la respuesta de las hembras a otros sonidos conocidos como modificadores de comportamiento, que es la canción "pre-copulatoria" cantada por machos apareados. De aquí que las aristas no son posiblemente los únicos órganos de recepción de la vibración de sonidos. Hembras vírgenes sin aristas son atraídas por feromonas producidas por machos y las aristas no muestran evidencia de "quemosensilla". Estos órganos parecen estar involucrados solamente como receptores mecánicos. Aparentemente, el canto de los machos de *A. suspensa* son producidos solamente por las alas, puesto que los niveles de la presión del sonido aumenta en proporción a la cantidad de superficie del ala, y no hay ningún rasgo morfológico que pueda hacer un mecanismo de estridulación hecho por la fuerza motriz del ala. Hay un pequeño dimorfismo sexual en la forma del ala (las alas de los machos son mas ovales). Comparaciones entre tefritidos con varias formas de alas y señales de cortejo, sugieren que el dimorfismo puede ser debido a modificaciones en el ala del macho para producir sonidos.

A few flies make sounds during courtship and territorial defense (Burk 1981). The best known are the "love songs" of *Drosophila* spp. (Bennet-Clark and Ewing 1970, see also contributions in Huettel 1985). However, there is information accumulating on the function and structure of acoustic signals in the Caribbean fruit fly, (caribfly) (*Anastrepha suspensa* [Loew])

(Diptera: Tephritidae). Caribflies produce 2 sounds in sexual context: the calling song, consisting of repeated pulse trains (bursts) broadcast from territories on host-plant leaves, and the precopulatory song emitted just prior to, and during insertion of the aedeagus (Webb et al. 1976). The calling song attracts females in field cages (Webb et al. 1983a) and increases the activity of virgin, but not of mated, females in the laboratory (Sivinski et al. 1984). Precopulatory song influences male acceptance by females and may be a means of advertising male vigor. Both loud volume and a continuous, non-pulsed structure increase the likelihood that a male will complete a coupling (Sivinski et al. 1984).

The means by which tephritid flies perceive these sexual messages have not been previously examined, though in another acalyptrate family, the Drosophilidae, sound is perceived through the vibration of the aristae stimulating Johnston's organ on the 2nd antennal segment (e.g., Manning 1967, Ewing 1978). We show here that the caribfly sounds are produced by the wings and some are received *via* the aristae, but that these antennal structures may not be the sole organs of hearing.

METHODS

SOUND RECEPTION

In order to determine the acoustic sensitivity of the aristae, adult flies were cold-anesthetized. Half of the anesthetized flies had at least 90% of their aristae removed with forceps, while the rest served as controls. Previous experiments had shown that virgin females respond to calling song with increased flight and walking (Sivinski et al. 1984). Differences in the response to sexual signals of virgin females with and without aristae were searched for in the following manner. Two observers, in an anechoic chamber, simultaneously measured activity in an experimental and in a control replication by counting the number of times 25 flies in a 20 cm x 20 cm x 20 cm screen wire cage crossed a line bisecting the cage.

Both cages had a speaker simultaneously emitting the same recorded calling song at a sound pressure level (SPL) (odB re 20 μ pa) of 55 dB at cage-center (note that due to meter inertia calling song SPLs are estimates rather than precise measurements). Counts were made during 6 alternating 5-min periods of broadcast and silence. Ten replicates were performed.

To determine if aristae might have additional chemosensory properties, 15 virgin ♀♀ with and 15 without aristae were placed in an olfactometer, a device that traps females moving in the direction of an odor stimulus. Males, substantially muted by wing removal, served as a pheromone source. Deaulation was an attempt to avoid confusion of signal modes, i.e., to provide both supposedly deaf and hearing females with a similar set of signals. Ten replicates were made.

EFFECT OF ARISTAE REMOVAL ON COPULATORY BEHAVIOUR

Precopulatory song intensity influences female reception of mounted males (Burk & Webb 1983, Sivinski et al. 1984). It seemed reasonable that if aristae removal deafened females, such flies might be less likely to allow penetration, or that males would have to sing more loudly, or for longer periods of time to overcome female resistance. We observed intact and

females without aristae enclosed individually with a male fly in a 7.5 cm x 7.5 cm screen wire cylinder. The number of rejections a male suffered, the time between introduction and copulation, and the duration of the precopulatory song were recorded.

Precopulatory song SPL was measured with a 12.7 mm Brüel & Kjaer Model 4145® condenser microphone held at 2.5 mm from the flies, and coupled through a cathode follower to a Brüel & Kjaer Model 2608® microphone amplifier and SPL meter. Note that the SPL of the relatively continuous precopulatory song can be measured without the problem of meter inertia that plagues attempts to quantify the periodic calling song.

THE EFFECT OF CHANGES IN WING AREA ON SOUND PRODUCTION

We clipped the wings of male caribflies resulting in losses of 40, 60, 85 and 95% of the wing area ($N_{100 \text{ percent}} = 119$, $N_{60 \text{ percent}} = 6$, $N_{40 \text{ percent}} = 7$, $N_{15 \text{ percent}} = 2$, $N_5 \text{ percent} = 3$). These percentages were determined by cutting a wing image projected by a microprojector onto a piece of paper. The resulting model was cut in the same manner as the wings and the fragments weighed. Clipped males were placed with virgin females and the SPL of their precopulatory songs measured in the previously described manner.

SEXUAL DIMORPHISM IN WINGS

If wings are organs of sound production, it might be expected that their structure would be specialized for singing in the more acoustical sex (males). The shape (length/width) of male and female wings were compared by determining the width of the wings measured through the cross vein that separates the 2nd dorsal and the 2nd basal cells, to the wing margins and the length of the distal portion of the wing from the junction of the previously mentioned cross vein with the M_{1+2} vein to the distal wing margin. Measurements were made by 2 methods, with a micrometer mounted in a stereomicroscope ($n \text{ } \delta \delta = 21$, $n \text{ } \varnothing \varnothing = 20$), and by microprojector that projected an enlarged image on a piece of paper ($n \text{ } \delta \delta = 26$, $n \text{ } \varnothing \varnothing = 26$) (see Sharp 1979). In order to search for allometric relationships between size and wing shape, the sizes of flies were determined in the micrometer-gathered data by measuring the length of a hind femur. The wings of another singing fruit fly *Toxotrypana curvicauda* Gerstaecker and a silent species *Rhagoletis pomonella* (Walsh) were measured with a micrometer so that our sound production hypothesis could be tested by comparison ($N_{TP} \delta = 21$, $N_{TP} \varnothing = 28$; $N_{RP} \delta = 10$, $N_{RP} \varnothing = 10$, see respectively Landolt et al. 1985 and Prokopy and Bush 1973 for details of courtship).

RESULTS

SOUND RECEPTION

Virgin female flies with aristae were more active during periods of calling-song broadcast than during alternating periods of silence (Wilcoxon paired test $P < 0.05$). There was no significant difference in the activities during sound and silence of females with their aristae removed ($P > 0.25$). The ability of flies without aristae to move about was not affected adversely by handling during the operation. Likewise, there was no difference in

the response of females with and without aristaе to the pheromones of mated males (28 with and 39 without aristaе were captured $P > .05$). Electronmicrographs of aristaе showed none of the surface features (e.g., pits) associated with chemoreception. When this lack of sensilla is coupled to the undiminished response to distance pheromone by females without aristaе, it seems that aristaе in caribflies are not chemosensitive and are primarily mechanoreceptors.

EFFECTS OF ARISTAE REMOVAL ON COPULATORY BEHAVIOUR

There was no indication that removal of aristaе influenced copulatory behavior. Females without aristaе were no more apt than intact females to reject mounted males (28% of 58 ♂♂ mounting intact females were rejected at least once, compared to 31% of 51 ♂♂ mounting aristaеless females). Nor did intact females accept males more rapidly (\bar{X} 11.9 min from introduction *vs.* 13.3 min $P = 0.42$), have longer matings (\bar{X} 41.9 min *vs.* 39.8 min $P = 0.49$), or have suitors that sang shorter songs (\bar{X} 71.8 sec *vs.* 55.9 sec $P = 0.41$).

THE EFFECT OF CHANGES IN WING AREA ON SOUND PRODUCTION

The sound pressure level of a male precopulatory song decreased with decreasing surface area of the wing, suggesting that the wing is either the agent of sound production or the power source for a sound producing organ ($r^2 = 0.88$, $P < 0.01$), note that the last clipping leaves ca. 5% of the wing producing ca. 6% of the original SPL). There is no evidence of morphological features that might make up a stridulatory organ and it thus seems likely that caribfly songs are sung only with the wings with perhaps some additional vibrations of thoracic structures (see Esch and Wilson 1967).

SEXUAL DIMORPHISM IN WINGS

The ratio of wing length to width is significantly greater in female flies, i.e., the wings of males are slightly "stubbier" (micrometer obtained ratios \bar{X} ♂ = 1.65, \bar{X} ♀ = 1.69, Wilcoxon rank-sum test $P = 0.02$, $P = 0.01$; microprojector \bar{X} ♂ = 1.68, \bar{X} ♀ = 1.72, $P < 0.01$). This is not due to an allometric relationship; that is, there is not a change of wing shape with size that would generate a *de facto* sexual dimorphism since males are generally smaller. In fact, there is an opposite relationship in female flies where the longer the femur the less the length of the wing relative to width ($r = -.57$, $P = 0.009$). There is no significant relationship between size and shape in males. In the papaya fruit fly, *T. curvicauda*, which also produces wing generated courtship sounds, male wings are again stubbier (\bar{X} ♂ = 1.90, \bar{X} ♀ = 2.13, $P < 0.0001$). However the sexually silent apple maggot, *R. pomonella*, has no sexual dimorphism in wing shape (\bar{X} ♂ = 1.60; \bar{X} ♀ = 1.59, $P = 0.64$). The greater surface area of male wings of a given length could be a means of displacing more air and hence producing a louder sound.

DISCUSSION

Removal of aristaе inhibited female response to calling song but does

not obviously affect coupling, a process known to be influenced by precopulatory song. Perhaps the simplest way of resolving these conflicting results is to posit at least 2 sites of sound/vibration reception. It is plausible that the sound produced by a male in physical contact could be perceived through mechanoreceptors on the female body, legs and/or wings. If so, perhaps the removal of organs adapted for sensing air displacement (i.e., aristae) would not significantly affect reception of precopulatory signals or subsequent female behavior.

Averhoff et al. (1979) have argued that the aristae of *Drosophila* bear chemosensory sensilla and that changes in behavior recorded in *Drosophila* after removal of antennae may not be due to deafness, but an inability to perceive pheromones. Both bioassay and micromorphological examination failed to find any evidence of chemosensitivity in caribfly aristae.

Evidence suggests that the caribfly sounds are solely or principally generated by wing movements. It is tempting to ascribe any differences in the wings of the sexes to modification for male sound production. However, sexual dimorphism in wing shape also might increase the signaling surface for visual displays, or reflect changes in wing loading evolved because of a dimorphism in flight activity. Comparing wing shape in tephritids with different signaling systems might help illuminate the functions of sexual dimorphism in wing shape. The reader will recall that male wings are more oval in the singing species *A. suspensa* and *T. curvicauda*. However, *R. pomonella*, which moves its wings in courtship but does not sing has no such sexual dimorphism. Male wings in the Mediterranean fruit fly, *Ceratitis capitata* (Wied.), another acoustic singer that produces both stridulatory and wing generated sounds are again stubbier than those of the female (Keiser et al. 1973, Webb et al. 1983b). Sharp (1979) has determined wing width and length using different measurements than ours for a number of tephritids. These can be used for cautious comparisons with our data (note that these data would show no sexual dimorphism in *A. suspensa*). *Dacus* spp. acoustic signals are due to stridulation (Monro 1953). *D. cucurbitae* (Coquillett) and *D. oleae* (Gmelin) have wing length/width ratios that are opposite *A. suspensa*, while *D. dorsalis* has no apparent sexual dimorphism. Thus the comparative evidence suggests that ovalness in male wings may have an acoustic function.

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